

CLAIMS

- 1 1. A process of detecting an oligonucleotide elongation, the process
2 comprising the steps of:
- 3 (a) providing an oligonucleotide;
- 4 (b) combining a detectable moiety and the oligonucleotide to form a
5 labeled oligonucleotide, the labeled oligonucleotide characterized
6 by an association independent of a dual contribution covalent bond
7 between the detectable moiety and the oligonucleotide;
- 8 (c) adding the labeled oligonucleotide to an oligonucleotide elongation
9 mixture;
- 10 (d) initiating an elongation reaction in the oligonucleotide elongation
11 mixture; and
- 12 (e) assaying for the labeled oligonucleotide in the oligonucleotide
13 elongation mixture to detect the oligonucleotide elongation.
- 1 2. The process of claim 1 wherein the non-covalent association is
2 selected from the group consisting of: an ionic bond, a hydrogen bond, a Van der
3 Waals interaction and an organometallic coordinate covalent bond.
- 1 3. The process of claim 1 wherein the detectable moiety comprises a
2 fluorophore.
- 1 4. The process of claim 1 wherein the detectable moiety comprises a
2 metal-containing fluorescent compound.
- 1 5. The process of claim 4 wherein the metal-containing fluorescent
2 compound comprises platinum.
- 1 6. The process of claim 4 wherein the metal-containing fluorescent
2 compound comprises a metal selected from the group consisting of: palladium,
3 rhodium, ruthenium, osmium, and iridium.

1 7. The process of claim 1 wherein the elongation reaction is a
2 polymerase chain reaction.

1 8. The process of claim 1 wherein the elongation reaction is a reverse
2 transcription reaction.

1 9. The process of claim 1 wherein the elongation reaction is a primer
2 extension reaction.

1 10. The process of claim 1 wherein the elongation reaction is a ligase
2 chain reaction.

1 11. The process of claim 1 wherein the process further comprises the
2 step of purifying the labeled oligonucleotide.

1 12. The process of claim 1 wherein the step of assaying the labeled
2 oligonucleotide comprises a measurement of fluorescence polarization.

1 13. The process of claim 1 wherein the step of assaying the labeled
2 oligonucleotide comprises a measurement of fluorescence intensity.

1 14. The process of claim 1 wherein the step of assaying the labeled
2 oligonucleotide comprises a measurement of fluorescence resonance energy
3 transfer.

1 15. A process of detecting an oligonucleotide elongation, the process
2 comprising the steps of:

3 (a) providing an oligonucleotide elongation reaction mixture
4 comprising an oligonucleotide labeled with a fluorescent
5 compound by an association independent of a dual contribution
6 covalent bond;

- 7 (b) measuring a fluorescence parameter in the oligonucleotide
8 elongation reaction mixture at a first time point to obtain a test
9 measurement; and
10 (c) comparing the test measurement with a reference measurement to
11 detect the oligonucleotide elongation.

1 16. The process of claim 15 wherein the reference is a second
2 **measurement of a fluorescence parameter in the oligonucleotide reaction mixture**
3 **at a second time point.**

1 17. The process of claim 16 wherein the second time point is before
2 initiation of the elongation reaction.

1 18. The process of claim 16 wherein the first and second time points
2 are after initiation of the elongation reaction.

1 19. The process of claim 15 wherein the reference is a measurement of
2 a fluorescence parameter in a second oligonucleotide extension reaction mixture.

1 20. The process of claim 15 wherein the non-covalent association is
2 selected from the group consisting of: an ionic bond, a hydrogen bond, a Van der
3 Waals interaction and an organometallic coordinate covalent bond.

1 21. The process of claim 15 wherein the fluorescent compound is a
2 metal-containing fluorescent compound.

1 22. The process of claim 21 wherein the metal-containing fluorescent
2 compound comprises platinum.

1 23. The process of claim 21 wherein the metal-containing fluorescent
2 compound comprises a metal selected from the group consisting of: palladium,
3 rhodium, ruthenium, osmium, and iridium.

1 24. The process of claim 15 wherein the elongation reaction is a
2 polymerase chain reaction.

1 25. The process of claim 15 wherein the elongation reaction is a
2 reverse transcription reaction.

1 26. The process of claim 15 wherein the elongation reaction is a primer
2 extension reaction.

1 27. The process of claim 15 wherein the elongation reaction is a ligase
2 chain reaction.

1 28. The process of claim 15 wherein the fluorescence parameter is
2 selected from the group consisting of: fluorescence polarization and fluorescence
3 intensity and fluorescence resonance energy transfer.

1 29. A process of detecting an oligonucleotide elongation, the process
2 comprising the steps of:

- 3 (a) providing an oligonucleotide elongation reaction mixture
4 comprising an oligonucleotide labeled with a metal-containing
5 fluorescent compound;
6 (b) measuring a fluorescence parameter associated with the metal-
7 containing fluorescent compound in the oligonucleotide elongation
8 reaction mixture at a first time point to obtain a test measurement;
9 and
10 (c) comparing the test measurement with a reference measurement to
11 detect the oligonucleotide elongation.

1 30. The process of claim 29 wherein the metal-containing fluorescent
2 compound comprises platinum.

1 31. The process of claim 29 wherein the metal-containing fluorescent
2 forms a coordinate covalent bond to label the oligonucleotide.

1 32. The process of claim 29 wherein the metal-containing fluorescent
2 compound comprises a metal selected from the group consisting of: palladium,
3 rhodium, ruthenium, osmium, and iridium.

1 33. The process of claim 29 wherein the elongation reaction mixture is
2 a polymerase chain reaction mixture.

1 34. The process of claim 29 wherein the fluorescence parameter is
2 selected from the group consisting of: fluorescence polarization, fluorescence
3 intensity and fluorescence resonance energy transfer.

1 35. The process of claim 29 wherein the reference is a second
2 measurement of a fluorescence parameter in the oligonucleotide reaction mixture
3 at a second time point.

1 36. The process of claim 35 wherein the second time point is before
2 initiation of the elongation reaction.

1 37. The process of claim 35 wherein the first and second time points
2 are after initiation of the elongation reaction.

1 38. The process of claim 29 wherein the reference is a measurement of
2 a fluorescence parameter in a second oligonucleotide extension reaction mixture.

1 39. A process of detecting formation of an oligonucleotide hybrid, the
2 process comprising the steps of:

3 (a) providing a hybridization reaction mixture comprising an
4 oligonucleotide labeled with a metal-containing fluorescent
5 compound;

- 6 (b) measuring a fluorescence parameter associated with the metal-
7 containing fluorescent compound in the hybridization reaction
8 mixture at a first time point to obtain a test measurement; and
9 (c) comparing the test measurement with a reference measurement to
10 detect the oligonucleotide hybridization.

1 40. The process of claim 39 wherein the metal-containing fluorescent
2 compound comprises platinum.

1 41. The process of claim 39 wherein the metal-containing fluorescent
2 forms a coordinate covalent bond to label the oligonucleotide.

1 42. The process of claim 39 wherein the metal-containing fluorescent
2 compound comprises a metal selected from the group consisting of: palladium,
3 rhodium, ruthenium, osmium, and iridium.

1 43. The process of claim 39 wherein the reference is a second
2 measurement of a fluorescence parameter in the oligonucleotide reaction mixture
3 at a second time point.

1 44. The process of claim 43 wherein the second time point is before
2 initiation of the elongation reaction.

1 45. The process of claim 43 wherein the first and second time points
2 are after initiation of the elongation reaction.

1 46. The process of claim 39 wherein the reference is a measurement of
2 a fluorescence parameter in a second oligonucleotide extension reaction mixture.

1 47. The process of claim 35 wherein the fluorescence parameter is
2 selected from the group consisting of: fluorescence polarization, fluorescence
3 intensity and fluorescence resonance energy transfer.

1 48. A process for detection of changes in a nucleic acid essentially as
2 described herein in any of the examples.

1 49. A process for nucleic acid quantification essentially as described
2 herein in any of the examples.

1 50. A commercial package comprising a metal-containing fluorescent
2 compound reaction mixture component along with instructions for use thereof to
3 detect changes in an oligonucleotide indicative of elongation or hybridization.

1 51. The use of a detectable moiety attached post-synthesis to an
2 oligonucleotide for real-time detection of changes in nucleic acid elongation,
3 amplification or hybridization.

1 52. The use of claim 51 wherein the detectable moiety is a fluorophore.

1 53. The use of claim 52 wherein the fluorophore is a metal-containing
2 fluorescent compound.

1 54. The use of claim 53 wherein the metal-containing fluorescent
2 compound contains platinum.